SUMMARY OF NEW AWARDS
BIOENGINEERING RESEARCH PARTNERSHIPS (BRP)
FY 2004

BECON

Grant:	1R01CA108467-01	
Principal Investigator:	ABBOTT, NICHOLAS L	PHD
Title:	Biomolecular Analysis using Liquid Crystals	
Institution:	UNIVERSITY OF WISCONSIN MADISON	MADISON, WI
Project Period:	2004/09/03-2008/08/31	

DESCRIPTION (provided by applicant): The University of Wisconsin Bioengineering Research Partnership (UW-BRP) will focus on the development of new molecular analysis tools that possess the potential to be used to identify and validate biological endpoints whereby the activity of novel anti-cancer agents can be more accurately and rapidly evaluated as to their molecular mechanism(s) and clinical relevance. The work will initially be focused on the analysis of the epidermal growth factor receptor (EGFR), given that its over expression and mutation has been well-associated with some of the most incurable cancers. However, it is anticipated that the principles to be developed will be sufficiently versatile to be applied to other key signaling molecules in the future. Whereas the basis of existing high throughput screens largely restricts their application to in vitro molecular analyses of enriched preparations of receptors or other signaling molecules, the UW-BRP seeks to establish principles for tools that can also be applied to the analysis of samples from cultured cells and from biopsies of xenographs and spontaneous tumor tissues. This capability will ultimately enable a fundamental approach that will span the molecular, cellular and tissue levels and will be used in both basic research and in animal and human clinical trials. In the present proposal, a multidisciplinary team of researchers with diverse expertise in chemical and biological engineering, chemistry and biochemistry, and the biomolecular and biomedical sciences proposes to develop a broadly-applicable bioanalytical approach that integrates advances in the following areas: a) the nano-fabrication of surfaces, b) the development of synthetic and biochemical strategies for the covalent and oriented immobilization of proteins and peptides on surfaces, c) the implementation of liquid crystals as highly sensitive reporters of the presence of proteins captured on surfaces, and d) the investigation of key cell signaling proteins that participate in processes associated with carcinogenesis. Specifically, the analytical characteristics of liquid crystals for reporting the behavior of the well-recognized anti-cancer target, i.e. the EGF receptor, will be compared to conventional analytical methods in a study that will a) rapidly and sensitively assess the levels and activity of wild-type and mutant human EGF receptor in biological samples, b) test the hypothesis that wild-type and oncogenic forms of the EGF receptor exhibit differential inhibitor specificity, and c) assess if agents that potently inhibit EGF-mediated events in vitro will also exhibit a capacity to antagonize EGF receptor expression and/or activity in cell culture. These studies will use the EGF receptor system as a prototype and it is anticipated that the technology will be readily adaptable to a wide range of other molecular targets. In the long term, these new tools should be useful for the assessment of the molecular mechanisms and consequences of anti-cancer agents, thereby facilitating their research from basic biology through to clinical assessment of efficacy.

Grant:	1R01HG003275-01	
Principal Investigator:	CERRINA, FRANCESCO	PHD
Title:	Light-directed synthesis of genes & other biomolecules	
Institution:	UNIVERSITY OF WISCONSIN MADISON	MADISON, WI
Project Period:	2004/08/09-2007/07/31	

DESCRIPTION (provided by applicant): This University of Wisconsin Bioengineering Research Partnership proposes to develop technology capable of rapid, inexpensive, and robust production of long dsDNA molecules (genes) up to 10 kilobases in length. The genes will be assembled from individual oligonucleotides synthesized in parallel on DNA arrays using the Maskless Array Synthesizer (MAS), which permits the production of custom DNA microarrays with 786,000 features in only 4 hours. Light-directed "safety-catch" release chemistry will permit desired array components to be released from the surface in sets to form specific sub-assemblies, which in turn will be assembled into the final long

dsDNA product. Biological error correction methods, based upon the ability of the mismatch-binding protein MutS to recognize and remove mismatched duplexes, will ensure high fidelity. All of these functions will be merged into an integrated system - the Automated Gene Synthesis (AGS). The ability to make complete genes on demand, inexpensively, and with rapid turn around time, has revolutionary implications for a wide range of biological and medical research. The UW Bioengineering Partnership is made up of an exceptional interdisciplinary team of researchers with expertise in the areas of engineering, chemistry, informatics, and molecular biology and cellular biology. Team members have a proven track record in developing cutting edge tools for biological research. The inclusion of NimbleGen Systems on the team, as an industrial partner, brings additional resources in chemistry and MAS technology to accelerate development. In addition to the primary focus of the group upon total gene synthesis, this bioengineering effort will also apply the massively parallel control of light to fuel basic research in the areas of combinatorial chemistry of small molecules and the detection of surface interactions. Biological relevance of the technologies will be ensured by means of three specific applications: the rapid low cost production of DNA for gene targeting in stem cells (all five of the cell lines to be used in this proposal (HI, H7, H9, H13, and H14) are all listed on the NIH registry), high-throughput protein synthesis, and high density on-chip SNP detection using surface invasive cleavage reactions.

Grant:	1R01HL073598-01A1	
Principal Investigator:	CORLEY, RICHARD A	PHD
Title:	3D IMAGING & COMPUTER MODEL OF THE RESPIRATORY TRACT	
Institution:	BATTELLE PACIFIC NORTHWEST LABORATORIES	RICHLAND, WA
Project Period:	2004/09/01-2009/08/31	

DESCRIPTION (provided by applicant): The respiratory tract is one of the main interfaces between the body and the environment. Its major structural components are designed to maximize gas exchange and provide sensory input (i.e. odor detection). As such, the respiratory tract can become a target for a broad range of airborne environmental agents contributing to an expansive array of human diseases. Alterations in the structure or function of the respiratory system by diseases can dramatically affect the interface with the environment and alter the quality of life. To improve our ability to predict the dosimetry and thus the consequences of airborne pollutants (gases, vapors, particulates or atmospheric releases of chemical/biological weapons) or drugs intentionally administered by inhalation for normal or potentially sensitive populations, 3-dimensional (3D), biologically based models of the respiratory tracts of animals and humans will be developed by a cross-disciplinary team of mathematicians, physicists, chemists, and biologists. The overall specific aims of this partnership are to: (1) develop and apply magnetic resonance imaging and fluorescent microsphere techniques to determine the dynamic, 3D structural and functional properties of the respiratory tract; (2) determine the 3D cellular organization and metabolic capacity; (3) develop and extend software and computational capabilities for 3D modeling and upscaling techniques for cellular-to-organ model integration; (4) develop a normalized atlas of rat geometries with explicit measures of variability; (5) conduct in vivo gas exchange and particulate dosimetry studies for model validation and identification of model uncertainties; and (6) provide a web-based "pulmonary physiome" platform for dissemination and training of researchers and clinicians in the use of imaging and annotated model databases. Five projects are designed to provide the necessary data on the dynamic structure and function of the respiratory system for the development and validation of the computational models. To support these five projects, three technology development cores will be established in advanced imaging, computation, and database/modeling access and training for external users. A fourth core will serve as an administrative interface and will provide statistical support among the participating institutions and projects.

Grant:	2R01HL064365-06A1	
Principal Investigator:	CRANDALL, EDWARD D	MD ENGINEERING:CHEMICAL
Title:	Absorption mechanisms for peptide/protein drugs via lung	
Institution:	UNIVERSITY OF SOUTHERN CALIFORNIA	LOS ANGELES, CA
Project Period:	1999/09/30-2009/07/31	

DESCRIPTION (provided by applicant): Oral administration of newly bioengineered peptide/protein drugs is often ineffective due to degradation by gastric and intestinal digestive enzymes. As an alternative route for systemic absorption of such protein/ peptide drugs, transpulmonary delivery has shown considerable potential. In this proposal, our long-term goals are to elucidate the mechanisms for absorption of various classes of peptide/protein drugs across the alveolar epithelium (that affords a vast surface area and relatively low protease activity). Although pulmonary

delivery of protein / peptide drugs in animal studies has been shown to yield much better bioavailability compared to oral delivery, absorption mechanisms and pathways are mostly undefined to date. Many bioengineering-related issues are associated with pulmonary drug delivery, including formulation of specific drugs, modes of delivery and transport mechanisms. Of these, we will investigate various transport mechanisms that facilitate absorption of peptide/protein drugs across alveolar epithelium, using cultured rat and human alveolar epithelial cell monolayers as in vitro models, and will extend key in vitro findings to in vivo rat lung studies. Model proteins/peptides to be explored range from oligopeptides to proteins of biological importance (e.g., calcitonin, insulin, granulocyte-colony stimulating factor, and human growth hormone). Our research plan is subdivided into three major projects: i) investigate transcellular transport mechanisms (e.g., fluid-phase, receptor-mediated and/or adsorptive transcytosis) for absorption of model drugs across the alveolar epithelial barrier, ii) elucidate strategies for enhancement of alveolar epithelial absorption of protein/peptide drugs via paracellular and/or transcellular routes (e.g., transient alteration of barrier properties), and iii) study enhanced receptor-mediated transcytosis of macromolecule drugs (e.g., conjugation with transferrin in the presence of trans-Golgi disruptors). The collaborative investigation of pulmonary protein/peptide drug absorption among several different biomedical research laboratories, utilizing different experimental approaches spanning cell biology to bioengineering/physiology, promises success in providing pertinent information on advancing practical approaches to pulmonary drug delivery.

Grant: 1R01GM066712-01A1

Principal Investigator: DORDICK, JONATHAN S.

Title: High-Throughput Solid-Phase Combinatorial Biocatalysis

Institution: RENSSELAER POLYTECHNIC INSTITUTE TROY, NY

Project Period: 2004/02/03-2008/01/31

DESCRIPTION (provided by applicant): Rapid developments in genomics, proteomics, and combinatorial chemistry have reshaped the field of drug discovery, providing new drug targets for selective screens and new compounds to be tested in those screens. While combinatorial methods have given rise to large libraries of compounds, typically these compounds result in improved lead candidates that must undergo further transformations by conventional medicinal chemistry to yield new drug candidates. Bioengineering, in the context of high-throughput combinatorial methodologies, has not impacted lead optimization nearly as much as it has lead discovery, mainly because of the highly selective, intricate chemistries often required to optimize lead compounds and the lack of a suitably broad high-throughput platform. Combinatorial biocatalysis can help overcome these obstacles by exploiting the exquisite selectivity and unique reactivity of enzymes and microbial biocatalysts; however, to date this technology has been limited to the derivatization of soluble substrates. We propose to expand the scope of combinatorial biocatalysis to include reactions on, and the generation of libraries from, lead molecules attached to solid and soluble polymer supports. In the process, we will develop a high-throughput, biocatalytic technology for drug discovery. The specific aims are: 1. To expand the breadth of biocatalysis on solid- and polymer-supported compounds in aqueous and nonaqueous media; 2. To develop strategies for attaching lead compounds and removing their derivatives from solid and polymeric supports; 3. To demonstrate high-throughput, combinatorial biocatalytic lead optimization of complex natural and synthetic molecules, screen resulting derivatives for biological activity, and scale up structurally and functionally interesting derivatives using biotransformations. A series of lead molecules will be used in this work, ranging from enzyme substrates that are attached onto solid and soluble polymer supports to complex compounds (the flavonoid bergenin and the current HIV-1 protease inhibitor indinavir). Successful completion of this research program will result in a powerful methodology that can be used by biomedical investigators in the search for new, more potent small molecule therapeutics.

Grant: 1R01CA103828-01

Principal Investigator: FERRARA, KATHERINE W PHD ELECTRICAL ENGINEERING

Title: Ultrasound Imaging and Local Drug Delivery in Tumors

Institution: UNIVERSITY OF CALIFORNIA DAVIS DAVIS, CA

Project Period: 2004/05/25-2009/04/30

DESCRIPTION (provided by applicant): Many promising studies indicate that ultrasound-enhanced delivery vehicles can be used to locally deliver a drug to a region of interest, with ultrasound imaging used to define the region to be treated and to monitor the inflow of the delivery vehicle. We will specifically explore the ultrasound-based mechanisms that allow new therapeutics to be concentrated in a user-specified region of interest within a tumor. These include the

use of ultrasonic radiation force to increase the capture of targeted delivery agents, the release of a compound through ultrasound-induced fragmentation of the vehicle, and the use of ultrasound to mechanically or chemically induce an increase in microvascular permeability. Drug delivery vehicles can be engineered to be manipulated by ultrasonic radiation force, where the ultrasound pressure deflects the flow of the agent to the vessel wall. The vehicles can also be coated with a targeting ligand, resulting in adhesion to endothelial cells. In addition, the vehicles can be designed such that ultrasound pressure produces fragmentation of a micron-sized sphere into particles on the order of tens to hundreds of nanometers, which are taken up more readily. UC Davis and Siemens Medical Systems will create an ultrasound system and transducers that can create an image with a typical clinical frequency (2.5-7.5 MHz) and disseminate a drug to a 3D region of interest with a lower center frequency (1 MHz). A full three-dimensional integrated system will be operational in year four of the proposed work, with prototypes available earlier. Although ultrasound-enhanced local delivery has shown great promise in vitro and in preliminary in vivo studies, we believe that a system that can specify a region of interest in three dimensions and monitor the inflow of the drug vehicle must be created in order to produce reproducible and quantitative improvements. UC Davis and ImaRx will cooperate in the development and evaluation of molecularly-targeted drug delivery vehicles, with new vehicles available beginning in the first year. Additionally, in the first two years of the project period, a team of UC Davis investigators will develop PET tools required to assess the biodistribution of the labeled agents with and without insonation, and these tools will be applied throughout this project and disseminated to the imaging community. With the advent of commercially-available systems for small animal imaging with PET, the development of techniques to label drugs and drug carriers should also be of great interest. We will initially develop these drug delivery strategies for the unique environment of tumors, although the techniques should have broad application.

Grant:	1R01HL074896-01A1	
Principal Investigator:	FUKAMACHI, KIYOTAKA	MD
Title:	Development and Clinical Testing of CorAide RVAD/BVAD	
Institution:	CLEVELAND CLINIC FOUNDATION	CLEVELAND, OH
Project Period:	2004/08/01-2004/08/02	

DESCRIPTION (provided by applicant): The use of implantable left ventricular assist devices (LVADs) has been increasing to serve the growing population of patients with end-stage congestive heart failure. However, up to 40% of patients have significant right ventricular (RV) failure that limits the utility of implantable LVAD therapy. RV failure leads to two problems: decreased forward flow and high right heart pressures that result in passive congestion of the liver, kidneys, and abdominal organs. Both factors contribute to multiorgan failure, the leading cause of death after LVAD implant. Such patients commonly require prolonged inotropic support or support with a right ventricular assist device (RVAD). Clinically available RVADs are not implantable devices and have several limitations due to poor blood compatibility, high infection rates, poor long-term durability, need for anticoagulation, need for a hospital stay, high mortality, and a less than ideal quality of life. We have reported a poor prognosis in patients with LVAD support who also required external RVAD support or prolonged inotropic support. A safe, effective, implantable RVAD could save the lives of many such patients with RV failure. We have developed the CorAide TM LVD-4000 Assist System, which is based on an implantable, third generation, centrifugal pump. A rotating assembly is fully suspended without mechanical contact or wear during operation. If the CorAide LVAD can be modified and used as an RVAD, the resulting CorAide biventricular ventricular assist device (BVAD) will be an ideal system for permanent support (destination therapy). The main objectives of this proposed program are to design, develop, and clinically evaluate an implantable RVAD that can be used as a component of an implantable BVAD for patients with severe biventricular failure. The specific aims are (1) Design and develop an implantable RVAD based on the CorAide LVAD, third generation centrifugal blood pump, (2) Design and develop an advanced fail-safe control algorithm capable of fixed speed or automatic mode that balances RVAD and LVAD performance, (3) Undertake in vivo characterization testing of the system both as an isolated RVAD and as a BVAD with the CorAide LVAD, (4) Undertake in vivo and in vitro reliability testing of the complete RVAD system, and (5) Obtain FDA approval for Investigational Device Exemption (IDE) and undertake clinical pilot studies using an institutionally approved program for patient selection and data collection. In this proposal, we will design and develop an RVAD in the first year, perform the characterization study in the second year, perform in vivo and in vitro reliability studies in the second and third years, and perform a clinical trial in the fourth and fifth years. The successful completion of this program will provide clinicians and patients with a safe and effective option for outpatient mechanical support that allows an excellent quality of life.

Grant:	1R01NS048826-01	
Principal Investigator:	GILLIS, KEVIN D	PHD

Title: Microchip devices to assay quantal exocytosis

Institution: UNIVERSITY OF MISSOURI COLUMBIA COLUMBIA, MO

Project Period: 2004/09/01-2009/07/31

DESCRIPTION (provided by applicant): The long-term objective of our research is to develop microdevices for high-throughput measurement of quantal exocytosis from neurons and neuroendocrine cells. Patch-clamp electrophysiological and carbon-fiber electrochemical approaches represent the state-of-the-art for high time resolution and high information content assays of exocytosis but are slow and labor-intensive. Biochemical assays of secretion from cell populations have limited time resolution and can not resolve individual quantal fusion events. We will use microchip technology to develop devices that can assay quantal exocytosis from thousands of cells in a day in order to greatly accelerate the pace of basic neuroscience research. This approach will also enable, for the first time, rapid and high information content screening of drug candidates that affect exocytosis of neurotransmitter. For example, L-DOPA used to treat Parkinson's disease acts by increasing the quantal content of dopamine release. The approach will be interdisciplinary and will bring together investigators with expertise is biomedical, electrical and mechanical engineering, materials science, physics, electrochemistry, physiology and biophysics. The specific aims are: 1) Develop approaches to automatically target individual cells to electrochemical microelectrodes on microfabricated devices. 2) Develop approaches to stimulate exocytosis from cells on microdevices including rapid microfluidic solution exchange, photolysis of caged Ca and electrical stimulation of action potentials. 3) Integrate new electrochemical electrode materials into microdevices to increase sensitivity and performance. 4) Develop electronic instrumentation to allow simultaneous recording of many channels of electrochemical or electrophysiological data. The five-year goal of the project is to have actual devices on the market to serve the exocytosis research community that are at least an order of magnitude faster than current carbon-fiber approaches. Our first-year milestones for each aim are: 1) Position one or more cells at predetermined sites on a microchip. 2) Exchange extracellular solution in < 100 ms on a microchip. 3) Characterize the electrochemical properties of a diamond-like carbon microelectrode. 4) Develop inexpensive modular circuitry for basic electrochemical measurements using off-the-shelf components that can be easily scaled for approximately 12 simultaneous channels.

Grant: 1R01HL073644-01A1

Principal Investigator: GILMOUR, ROBERT F

Title: MEMS Sensors for Arrhythmia Detection and Interventions

Institution: CORNELL UNIVERSITY ITHACA ITHACA, NY

Project Period: 2004/09/15-2009/08/31

DESCRIPTION (provided by applicant): Despite decades of intensive investigation, sudden death secondary to ventricular fibrillation (VF) remains a leading cause of mortality in the US and other developed countries. Recently, several promising hypotheses regarding the mechanism for VF have been introduced. However, it has not been possible using currently available experimental techniques to determine which theory (or theories) is most applicable to VF. To address this issue, we propose to: 1) construct a cardiac mapping system from nanofabricated components that is capable of assessing cardiac activation and repolarization with high spatial and temporal resolution and with minimal tissue damage; 2) use a novel phase mapping technique to analyze the mapping data, with the objective of identifying the location and number of phase singularities during sinus rhythm, ventricular tachycardia and VF; 3) use the phase singularity data to distinguish between three putative mechanisms for VF - an anchored rotor with fibrillatory conduction, a meandering rotor or multiple rotors. MEMs technology will be used to construct microscale mechanical needle-like structures with integrated electrodes that are ultrasonically activated, to minimize tissue damage during insertion. The electrode arrays will be used to map activation and repolarization in canine ventricular myocardium in vitro and in normal and acutely ischemic pig hearts in situ during fixed pacing and during VF. The mapping data will be analyzed using a fast Fourier-demodulation technique to identify singularities and wave vectors during VF. Computer models of 2- and 3-D myocardium also will be used to generate surrogate data sets for testing the analysis algorithms. The results of this study will lead to significant advances in three key areas: development of devices to map cardiac electrical activity with unprecedented spatial resolution; application of newer and more sophisticated techniques to analyze large mapping data sets; interpretation of high resolution mapping data within the context of novel hypotheses regarding the genesis of ventricular tachycardia and fibrillation. Taken together, these advances in data acquisition, analysis and interpretation are expected to lead to new and more effective means of identifying and treating patients at risk for the development of lethal ventricular tachyarrhythmias.

Grant: 2R01HL064795-06

Principal Investigator:	HALPERIN, HENRY R	MD
Title:	Magnetic Resonance Guided Electrophysiology Intervention	
Institution:	JOHNS HOPKINS UNIVERSITY	BALTIMORE, MD
Project Period:	1999/09/30-2009/05/31	

DESCRIPTION (provided by applicant): Ventricular tachyarrhythmias and atrial fibrillation occurring in patients with structurally abnormal hearts are the most important arrhythmias in contemporary cardiology, and despite much progress, remain therapeutic challenges. Invasive electrical studies of the heart (electrophysiologic studies) are often used in the diagnosis and therapy of arrhythmias, and many arrhythmias can be cured by selective destruction of critical electrical pathways with radiofrequency (RF) catheter ablation. A major limitation in studying arrhythmias in patients, however, is the lack of ability to accurately correlate anatomical and electrical information. Another major limitation is the lack of ability to visualize ablated areas of myocardium during catheter ablation procedures, making it difficult to confirm the presence of ablated lesions in the desired locations. We are developing ways of combining the anatomic information from magnetic resonance imaging (MRI), with electrophysiologic testing and ablation. We hypothesize that MRI, with MRI-compatible (non-magnetic) electrode catheters, catheter-tip location sensors, intracardiac receivers, real-time MRI scanner control, remote-control catheter manipulators, and 3- dimensional imaging software can (1) provide the ability to accurately visualize cardiac anatomy, (2) provide accurate navigation of catheters without radiation, (3) provide the ability to visualize ablated lesions, and (4) aid in producing more accurate electrical maps. Our previous project dealt with (1) technology development, (2) demonstration of the feasibility of MRI guidance of catheters in animals, and (3) lesion visualization in animals, and in patients with atrial arrhythmias. This competing continuation deals with (1) additional technology development, (2) improved integration of the different subsystems, (3) study of the determinants of successful ablation in patients undergoing standard ablations, and (4) broadening of the applications to real-time MRI guided therapy in patients with atrial and ventricular arrhythmias. The technologies developed in this project, should, in addition, be applicable to using MRI to guide interventional procedures in general. This project is a partnership between the Johns Hopkins University School of Medicine (Medicine, Radiology, and Biomedical Engineering), Robin Medical Inc., MicroHelix Inc., NaviCath Inc., and Irvine Biomedical, Inc. All entities have supplied resources to the project, and will continue to share the costs of the project. The School of Medicine has an ongoing commitment to developing cardiac MRI, as demonstrated by its substantial investment in MRI scanners, including one adjacent to the cardiac catheterization laboratory. These scanners have a magnet that is short enough to allow access to the groin vessels for placement of catheters for diagnostic and interventional procedures. Robin Medical has developed technologies for precisely localizing the tip of a catheter inside an MRI scanner, and is developing technology for deflecting the tip using the MRI magnetic fields. MicroHelix is developing specialized catheter electrodes that reject MRI electromagnetic interference. NaviCath is developing an MRI-compatible system for remote manipulation of catheters that will allow catheters to be manipulated in patients in MR scanners that are too long to allow easy access to the groin vessels. Irvine Biomedical is supplying non-magnetic electrode catheters for use in the MRI scanner.

Grant:	1R01EB000993-01A1	
Principal Investigator:	JACOBS, RUSSELL E	
Title:	Multimodal mPET & mMRI Imaging Instrumentation	
Institution:	CALIFORNIA INSTITUTE OF TECHNOLOGY	PASADENA, CA
Project Period:	2004/08/01-2009/07/31	

DESCRIPTION (provided by applicant): We propose to combine the best features of Positron Emission Tomography (PET) and Magnetic Resonance Imaging (MRI) modalities in a single instrument that will simultaneously record data in both imaging modalities. Moreover, we will develop labeled probes that can be detected in both PET and MRI to aid in the interpretation of complex biological processes. This system will be dedicated to the study of small animal model systems at the highest spatial and temporal resolutions attainable. We will build a high resolution, relatively high sensitivity multi-slice muPET scanner integrated within a customized 7T/30cm small animal MR system that will simultaneously record MR and PET images Through the use of fiber-optic couplings, the muPET system will interfere minimally with the muMR system, enabling high quality muPET and IJMRI data to be acquired essentially simultaneously and in near-perfect spatial registration. This system is a natural extension of earlier proof-of-principle systems and a newer prototype animal muPET system now nearing completion. The basic design elements of the system have been tested and demonstrated to work. The combined system we propose adds a number of important features to improve performance and ease of use for in vivo imaging studies. It also incorporates, for the first time, a multi-slice muPET system, with four detector rings simultaneously providing seven imaging planes, spanning an axial

field of view of approximately 8mm, with at least 2mm resolution. Simultaneous muPET/muMRI recordings will provide important correlations not available from temporally and spatially separate scans (e.g. BOLD MRI compared with FDG PET). The melded system will provide high resolution anatomical reference systems for muPET studies The 'in register'muMR images will be used to compute scatter and attenuation in the IJPET images and to estimate partial volume errors in the PET scans, thus aiding quantification of the PET signal. This system will open up a number of opportunities not possible with current independent technologies Among them are: Time correlated IJPET and MRS studies of drug distributions; cardiac, CNS and tumor cell metabolism. Simultaneous fMRI and muPET neuroreceptor brain mapping studies in small animals. Validation of new MRI probes using their PET counterparts. Dual PET/MRI labels will allow for "zooming-in" the MRI data collection scheme to those regions of the specimen containing the label, as well as providing for precise registration of the PET & MR images.

Grant:	1R01CA108468-01	
Principal Investigator:	NIE, SHUMING	PHD
Title:	Nanotechnology Linking Biomarkers with Cancer Behavior	
Institution:	EMORY UNIVERSITY	ATLANTA, GA
Project Period:	2004/09/15-2009/08/31	

DESCRIPTION (provided by applicant): This BRP application aims to establish a highly collaborative and multidisciplinary cancer nanotechnology program by integrating the bioengineering strengths of Georgia Tech (Atlanta, GA), the spectral imaging expertise of CSRI (Woburn, MA), and the cancer biology and clinical oncology experiences of Emory University School of Medicine (Urology, Pathology, Radiation Oncology, Winship Cancer Institute, and the VA Hospital, Atlanta, GA). With faculty participation from eight science, engineering, and clinical departments, and advised by a prominent Scientific Advisory Board (SAB), this Partnership incorporates broad expertise in bioengineering, bioinformatics, tumor biology, bioanalytical chemistry, systems biology, hematology / oncology, pathology, and urology. Its broad and long-term goal is to develop biomedical nanotechnology, biomolecular engineering, and bioinformatics tools for linking molecular signatures (biomarkers) of cancer and the host microenvironment with cancer behavior and clinical outcome. The proposed research is broadly applicable to many types of malignant tumors such as breast cancer, colorectal carcinoma, and lymphoma, but a particular focus will be placed on the biological behavior of human prostate cancer and its clinically lethal phenotypes. A compelling reason for this focus is that prostate cancer presents a number of unique challenges and opportunities in human oncology. Its widespread occurrence (about 220,000 new cases this year in the US), tendency for a long natural history, highly heterogeneous and multi-focal histopathology, and progression to hormone independence are still poorly understood. Faced with this reality, we propose to develop advanced nanoparticle technologies (e.g., molecular beacons, semiconductor quantum dots, and enhanced Raman probes) for ultrasensitive and multiplexed profiling of biomarkers on intact cancer cells or tissue specimens. In contrast to current molecular-profiling technologies, the use of encoded nanoparticle probes allows a seamless integration of traditional pathology and cancer biology with sensitive molecular analysis, a central theme that runs across the entire proposed research. Underlying this BRP is a strong track record of the senior investigators who have worked together successfully in attracting joint research grants. In addition, the Department of Biomedical Engineering, which was jointly established in 1997 by Georgia Tech and Emory University, has presented an unusual opportunity for research collaboration to bring bioengineering technologies and discoveries into medicine and vice versa. If funded, this cancer nanotechnology program will be housed in the Winship Cancer Institute, a new 280,000 sq ft cancer research and care building located on the Emory Campus and with a truly outstanding environment for collaborative and translational cancer research. In additional to basic knowledge on cancer biology and biomarkers, this Partnership is expected to yield at least three practical outcomes: (a) a database linking molecular signatures with cancer biology and clinical outcome, (b) bioconjugated nanoparticles for molecular profiling of cancer, and (c) muiltiplexed spectral imaging microscopes and software.

Grant:	1R01EB003824-01	
Principal Investigator:	NOLAN, JOHN P	PHD
Title:	Raman Flow Cytometry for Diagnostics and Drug Discovery	
Institution:	LA JOLLA BIOENGINEERING INSTITUTE	LA JOLLA, CA
Project Period:	2004/09/01-2009/08/31	

DESCRIPTION (provided by applicant): The ability to make quantitative, high throughput molecular measurements of biological systems is a critical need for many areas of biomedical research. This Bioengineering Research Partnership

(BRP) aims to develop a powerful new analytical platform for high throughput screening and selection based on Raman Flow Cytometry. This Partnership will develop new analytical instrumentation, optically encoded polymer resins for chemical synthesis and screening, and nanostructured materials with unique optically properties for sensitive reporting and encoding. The new technology will perform Raman spectroscopy on single particles in flow to enable new applications in sensitive multiplexed detection, drug discovery, and diagnostics. The Raman Flow Cytometry instrumentation, and applications will be developed by a Partnership involving engineers, biologists, and chemists from academia, government and industry. In the first year of the Partnership, we will modify a commercial particle sorter to detect individual Raman vibrational bands from single particles and sort these particles based on their optical signature. In Years 2-5, we will develop the ability to collect and analyze complete Raman spectra from single particles. In parallel, the Partnership will develop new encoding and reporting strategies for multiplexed molecular analysis and separation. This Raman Flow Cytometry technology will be applied to the development of therapeutics and diagnostics for bacterial pathogens and their toxins. Raman Flow Cytometry will be an important and general new analytical and separation capability that will impact many areas of basic and applied biomedical research in addition to the applications proposed here.

Grant:	1R01HG002644-01A1	
Principal Investigator:	QUAKE, STEPHEN R	PHD
Title:	Ultrasensitive Nanofluidic Devices for Genomic Analysis	
Institution:	CALIFORNIA INSTITUTE OF TECHNOLOGY	PASADENA, CA
Project Period:	2004/06/05-2009/04/30	

DESCRIPTION (provided by applicant): This partnership will bridge the technology development efforts of Caltech faculty in Applied Physics and Bioengineering with clinical needs of faculty at the USC Keck School of Medicine. We will apply technology for nanofluidic chips that has been developed at Caltech to problems of biological and medical interest. Using the combined resources of the Applied Physics and the Bioengineering programs, we will develop new nanotechnology to solve a number of bioengineering obstacles that presently exist for single cell genomic analysis on a chip. We will develop microfabricated chips with the ability to manipulate nanoliters of fluid; these chips will be used to perform highly parallel biochemical manipulations and genetic analyses of rare populations of cells. The chips will be able to create unique reagents that can be analyzed using conventional functional genomics techniques. A chip foundry will be created in order to produce research quantities of these chips that can be shared among collaborators. The chip technology will be used to investigate the following two problems of particular medical importance: factor and marker discovery in haematopoietic stem cells, and the discovery and characterization of unculturable pathogens in the human gut.

Grant:	1R01HG002647-01A1	
Principal Investigator:	RAMSEY, JOHN M	PHD
Title:	Nanotechnology for the Structural Interrogation of DNA	
Institution:	UNIVERSITY OF NORTH CAROLINA CHAPEL HILL	CHAPEL HILL, NC
Project Period:	2004/09/30-2006/08/31	

DESCRIPTION (provided by applicant): We propose a research program to achieve the goal of sequencing of single molecules of polynucleotides using conductance probes within a molecular scale aperture and to demonstrate the technical feasibility of this promising approach. There have recently been intriguing suggestions about how one might rapidly determine the sequence of a single DNA molecule contained in a buffer solution by transporting it through a voltage-biased nanoscale aperture while monitoring the ionic current through that aperture [Kasianowicz, 1996; Deamer, 2000]. Some suggestive proof-of-principle experiments have been demonstrated using lipid bilayer supported protein pores and observing variations in pore axial conductance. We contend that for this strategy to become a realizable technology, robust nanometer scale apertures must be fabricated using a combination of top-down and bottom-up approaches. In addition, interesting variants of this approach such as incorporating laterally opposed nanoelectrodes in a nanochannel for probing monomeric variations in the electrical properties of polynucleotides can only be achieved through nanofabrication. Our specific aims are listed below. Develop fabrication capabilities that combine top-down and bottom-up strategies for forming fluidic channels and electrical probes with length scales approaching 1 nm. Investigate the dependence of the length scale probed on nanopore axial and lateral dimensions. Compare the signal-to-noise ratio for axial and lateral conductance probes of single DNA strands. Determine variation of measurement signal-to-noise ratios as a function of chemical and physical parameters such as aperture size, buffer

conditions, interfacial hydrophobicity, and electrode size. Determine impact of polymer dynamics on fundamental limits of DNA structural determinations. Demonstrate proof-of-principle single molecule sequencing of polynucleotides based on achievement of these specific aims.

Grant:	1R01EB001719-01	
Principal Investigator:	RANSOM, JOHN	BA
Title:	Single Cell Multiplex Screening of Protein Families	
Institution:	NOVASITE PHARMACEUTICALS, INC.	SAN DIEGO, CA
Project Period:	2004/09/15-2008/08/31	

DESCRIPTION (provided by applicant): This proposal seeks to develop a unique high throughput screening (HTS) system composed of several integrated existing and novel automated components that will be uniquely capable of screening entire protein target families simultaneously. The following advantages will be realized due to its maximized efficiencies: i) improved drug selectivity and a reduction in drug side effects, and ii) greater than ten fold reduction in the cost of screening and drug candidate optimization. Additionally, novel instrumentation and methodologies will become available to the biomedical research community for academic and industrial use, including a high speed, six laser flow cytometer developed through DakoCytomation the (BRP partner) and an automated tissue culture system developed through The Automation Partnership. The Specific Aims are i) to assist in development of and procure an automated tissue culture system capable of handling over 300 microwell plates and nearly 2,000 distinct cell lines, ii) to expand the capabilities of our automated sampling system for flow cytometry (FCM) for greater speed and to enable unique analysis and sorting modes, iii) to develop at least 150 GPCR target cell lines through novel transfection and expression techniques and cellular evolution techniques, iv) to develop a single cell HTS screening platform based on six-laser FCM, multiparametric detection technology and an automated cell preparation system, v) to perform a focused library test screen of 10,000 compounds against the 150 GPCR targets, and vi) to develop the database and bioinformatics tools to integrate and help interpret the data. When fully integrated, tested and validated the system will be applicable to many target families of relevance to biomedical and pharmaceutical research, such as other receptor classes, and will have a broad range of drug discovery applications.

Grant:	1R01CA103830-01	
Principal Investigator:	RICHARDS-KORTUM, REBECCA R.	PHD MEDICAL PHYSICS
Title:	Optical Systems for In Vivo Molecular Imaging of Cancer	
Institution:	UNIVERSITY OF TEXAS AUSTIN	AUSTIN, TX
Project Period:	2004/08/01-2009/07/31	

DESCRIPTION (provided by applicant): Cancer is a major public health problem. Currently, classification of cancer is based on phenotypic markers. The identification of unique molecular markers of cancer has led to development of new molecular cancer therapies. Movement toward a molecular characterization of cancer would have important clinical benefits, including (1) detecting cancer earlier, (2) predicting risk of precancerous lesion progression, (3) detecting margins in the operating room in real time, (4) selecting molecular therapy rationally and (5) monitoring response to therapy in rear time at a molecular level. Imaging the molecular features of cancer requires molecular-specific contrast agents which can safely be used in vivo as well as cost-effective imaging systems to rapidly and non-invasively image the uptake, distribution and binding of these agents in vivo. Radiographic imaging modalities such as CT and MRI, although useful for delineating the deep extent of advanced carcinomas, are not sufficiently sensitive to detect small, intraepithelial lesions. Optical imaging is a new modality which enables real time, high resolution imaging of epithelial tissue. Optical imaging systems are inexpensive, robust and portable. Optical imaging systems are ideally suited for early detection of intraepithelial disease and to assess tumor margins and response to therapy. The goal of this proposal is to integrate development of optical imaging systems and contrast agents with advances in functional genomics. We will develop molecular-specific, optically active contrast agents that can be applied topically. We will also develop inexpensive, rugged and portable imaging systems to monitor the three-dimensional profile of targeted biomarkers. These contrast agents and imaging systems will have broad applicability to many types of cancer; here, we will develop and test agents and imaging systems for the cervix, oral cavity and the lung, which represent more than 20% of both tumor incidence and mortality worldwide. We will test the safety and efficacy of these contrast agents and imaging systems in animal models, providing data to support phase I and II clinical trials. The aims of this proposal are to: (1) Develop optically active contrast agents to target four molecular signatures of neoplasia, including EGFR, MMP, telomerase and alpha v integrin; (2) to identify promising new biomarkers for which contrast agents will

be developed using SAGE libraries, and to identify promising molecular probes for novel contrast agents using combinatorial methods; (3) to develop inexpensive, portable optical systems to image the morphologic and molecular signatures of neoplasia noninvasively in real time; and (4) to test these agents, delivery formulations and imaging systems in living biological systems of progressively increasing complexity. (5) Our final aim is to integrate these studies to develop a miniature imaging system, which when coupled with the contrast agents developed here, can be used for real time, molecular detection of neoplasia and to monitor, at the molecular level, whether a lesion is responding to therapy.

Grant:	1R01EB003806-01	
Principal Investigator:	STUPP, SAMUEL I	
Title:	Regenerative Scaffold Technologies for CNS and Diabetes	
Institution:	NORTHWESTERN UNIVERSITY	EVANSTON, IL
Project Period:	2004/09/04-2009/08/31	

DESCRIPTION (provided by applicant): Regenerative medicine is one of the great biomedical challenges of this century, seeking to regenerate parts of the human body throughout life lost to trauma, disease, or genetic factors. Real progress will hinge on our ability to combine effectively the frontiers of technology, biology, and clinical medicine to develop regenerative strategies. This Bioengineering Research Partnership (BRP), proposed by a team of seven investigators in the fields of neurology, surgery, endocrinology, materials science, chemistry, biomedical engineering, and chemical engineering, focuses on two specific challenges of great clinical importance, regeneration of the central nervous system (CNS) and cell replacement therapies for diabetic patients. In this application the target of the team is to develop multiple scaffold technologies and use CNS regeneration and pancreatic tissue replacement as their testing ground. The CNS targets include injection of self-assembling molecules and genetically engineered stem cells into the injured spinal cord or brain following stroke, and the diabetes targets include the development of a subcutaneous islet transplant. The four basic technologies are self-assembling nanofibers customizable to bear multiple tissue specific biological epitopes or have programmable delivery of growth factors; microporous biodegradable scaffolds that deliver genes or growth factors and guide cell migration; post-translationally modified recombinant polypeptides with customizable architecture and bioactivity; and enzyme-driven liquid-to-solid transitions of soluble bioactive peptides. The integrated scaffold technologies proposed include, the use of self-assembling nanofiber technology to modify microporous materials and create micro-nano hierarchical scaffolds, the adaptation of recombinant polypeptides for in situ enzyme driven solidification, and the development of bioactive two-phase molecular composite scaffolds containing linear polypeptides and peptide nanostructures.

Grant:	1R01GM071345-01	
Principal Investigator:	TONER, MEHMET	PHD
Title:	Cellular Engineering for Metabolic Stasis	
Institution:	MASSACHUSETTS GENERAL HOSPITAL	BOSTON, MA
Project Period:	2004/08/01-2008/07/31	

DESCRIPTION (provided by applicant): With the advancements being made in tissue engineering, cell transplantation, stem cell biology, and gene therapy, the clinical demand for effective long-term storage methods for cells and tissues will continue to increase. We propose to develop novel methods to biostabilization of mammalian cells for long-term preservation in a desiccated state at ambient temperature. In nature, many animals and organisms down regulate their metabolism and may enter into a state of stasis by either desiccation through removal of water from their cells (i.e., anhydrobiosis) or by a developmentally programmed arrest under full hydration (i.e., diapause). The ability to enter diapause prior to desiccation is crucial for the survivorship of many organisms that undergo natural states of dormancy. Furthermore, a common theme is that desiccation-tolerant animals accumulate large amounts of disaccharides, especially trehalose and sucrose. These sugars provide protective effects by forming stable sugar glasses at high water contents, and by stabilizing biological membranes and proteins through direct interaction with polar residues. We, therefore, hypothesize that metabolic preconditioning of mammalian cells to induce diapause-like state followed by controlled drying, storage, and rehydration conditions (i.e., physicochemical, biochemical, and metabolic) can be used to achieve desiccation tolerance in mammalian cells and tissues. To this end, our 3 distinct, but interactive, specific aims are: 1) To develop optimal physicochemical conditions to stabilize desiccated cells. 2) To metabolically precondition mammalian cells to improve survivorship during storage. 3) To develop metabolic and biophysical strategies to accelerate recovery of desiccated cells. This project is one of the first attempts to apply

engineering and quantitative concepts to achieving anhydrobiotic state in mammalian cells. It provides a systems view of the metabolic and cellular changes a cell encounters before, during, and after desiccation. This project is inspired by nature and it uses engineering concepts and approaches to translate nature's solution to long-term storage or "suspended animation" for mammalian systems. The proposed studies will significantly impact on human health by providing a solution to the problem of providing long-term storage of blood cells, stem cells, tissue engineered products, and cell-based biosensors for use in regenerative medicine, tissue engineering, and bioterrorism. In the short term, it will help increase the treatment modalities available to liver failure by providing stable, long-term stabilized cells for bioartificial liver assist devices. The longer-range outcome of the proposed research is to translate the information gained from these studies into whole organ preservation.

Grant:	1R01HL073646-01A1	
Principal Investigator:	WICKLINE, SAMUEL A	MD CARDIOLOGY
Title:	Methods in Molecular Imaging and Targeted Therapeutics	
Institution:	WASHINGTON UNIVERSITY	ST. LOUIS, MO
Project Period:	2004/07/15-2009/06/30	

DESCRIPTION (provided by applicant): The broad subject of this Biomedical Research Partnership (BRP) application is the development of novel multidimensional nanotechnologies for sensitive and specific imaging of molecular epitopes that are etiologic for atherosclerosis. The unifying hypothesis is that targeted molecular imaging with novel paramagnetic perfluorocarbon emulsion nanoparticle contrast agents can delineate selected molecular features of atherosclerotic lesions that are critical determinants of early lesion growth and later lesion instability. Noninvasive and early detection of these situations could enhance patient management and potentially reduce the incidence of myocardial infarction and stroke. The long-range goal is to produce a targeted nanoparticle contrast agent characterized by: 1) flexible targeting options depending on the binding ligand selected, 2) flexible imaging choices based on contrast mechanism best suited to the pathology in question, and 3) flexible opportunities for local delivery of therapeutic agents coupled directly with image-based quantification of local nanoparticle deposition. The technology is expected to enable early noninvasive detection of a variety of pathologies, convenient serial outpatient evaluation, and site-targeted delivery of therapeutics as clinically indicated. Stable and safe self-assembling nanoparticles will be developed, refined, and tested for visualization of pathological epitopes with the use of magnetic resonance imaging (MRI). Corporate partners who are involved in the research and intended commercialization are Kereos, Inc., Philips Medical Systems, Bristol-Myers Squibb Medical Imaging, and Dow Chemical.

Grant:	1R01CA108449-01	
Principal Investigator:	WONG, JOHN W	PHD BIOPHYSICS, OTHER
Title:	An Image Guided Small Animal Radiation Research Platform	
Institution:	WILLIAM BEAUMONT HOSPITAL RESEARCH INST	ROYAL OAK, MI
Project Period:	2004/04/01-2008/03/31	

DESCRIPTION (provided by applicant): Significant advances have been made in medical anatomical imaging over the past 25 years. More recently, there has been increased interest in small animal biological imaging research based on the increasing understanding of various molecular and genetic signals in tumor cells in vitro and in vivo. These initiatives have focused primarily on therapeutic research with drugs and other systemic agents, somewhat ironically, overlooking radiation therapy. The addition of biological imaging information to radiation therapy will have a major impact on the management and evaluation of treatment. Unfortunately, despite the interest in biological imaging research, animal radiation research methodology lags far behind clinical practices. Advanced techniques such as conformal, intensity modulated radiation therapy (IMRT) is increasingly becoming routine in human clinics, and has led to a shift in the clinical paradigm of the uniform dose delivery towards non-uniform dose delivery, particularly to the critical organs. The advent of new image guidance methods for short course radiation treatment will yet lead to the delivery of dose distributions of even greater nonuniformity. Regrettably, present laboratory research equipment prohibits testing of these paradigms in animal models. As a consequence, the advanced treatment technologies are applied clinically without any guidance from small animal radiation experimentation to evaluate efficacy in a preclinical setting. In this proposal we request funding for the de novo construction, testing and evaluation of an image guided small animal radiation research platform (SARRP) that will accurately deliver complex ionizing radiation dose distributions in small animal tumor model systems, mice, rats and rabbits. Specifically we propose 1) the construction of a gantry system containing three kilovoltage (kV) radiation sources that will have energy deposition resolution of

about 1mm and on-board cone beam CT imaging resolution of about 0.5 mm; 2) the development of dosimetric and treatment planning methodologies that parallel that for human treatment. Finally (3) the third specific aim is to develop methods of precise animal setups and to validate the imaging and irradiation capabilities of the system for accurate delivery of localized dose distributions in small animals. The research requires integration of expertise in mechanical engineering, x-ray optics and radiation dosimetry physics. The collaborative efforts are best coordinated in a Bioengineering Research Partnership (BRP). An Oversight Group is formed to evaluate the progress of the BRP, and to help identify opportunities and hypotheses for future research.